



Evaluation of the Physico-chemical and Antioxidant Activity Properties of Attieke Flour Enriched with Cashew Kernel (*Anacardium occidentale* L.) and Moringa (*Moringa oleifera* L.) Powders

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Authors' contributions

This work was carried out in collaboration among all authors. Author SK wrote the protocol, performed the various protocol analyzes and statistics also writes the manuscript. Author DS gave part of the reagent for carrying out the analyzes. Authors DS and EKK followed all the analyzes and corrected the manuscript. All authors read and approved the final manuscript.

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ABSTRACT

The objective of this study is to produce an infant flour based on attiéké enriched with *Moringa oleifera* and cashew kernel (*Anacardium occidentale* L.) powders. For this purpose, *Moringa oleifera* powder is incorporated in proportions of 10%, 15% and 20% into two types of composite flours (attiéké / unfermented cashew kernel and attiéké / fermented cashew kernel). Mineralogical, physico-chemical and antioxidant activity are performed. The results of the mineralogical analyzes showed potassium is the majority in composite flours. In addition, the calcium, magnesium, copper and zinc contents in the composite flours conformity with the standards. The fortification of attiéké flour with *Moringa oleifera* and cashew kernel powders resulted in an increase in phenolic

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compounds, antioxidant activity, as well as protein contents which vary from 11.59 ± 0.10 g / 100 g of 16.59 ± 0.26 g / 100 g. Moringa and cashew kernel powders have increased the nutritional quality of attiéké flour.

Keywords: Nutritional quality; attiéké; *Anacardium occidentale* L.; *Moringa oleifera*; fortification.

1. INTRODUCTION

Cassava (*Manihot esculenta*, Crantz) is an important staple food crop for millions of people in the tropics of the world. Cassava roots provide up to a third of daily calories [1], and contain mainly carbohydrates, of which 80% is starch [2]. Traditionally, cassava roots are processed by several methods, according to local customs and preferences [3] such as attiéké, gari, fufu, flour, fries, starch, syrups, dextrins and alcohol [4].

In Ivory Coast, attiéké is the most widely consumed cassava by-product in urban areas [5]. It constitutes approximately 5% of the food expenses of many Ivorian populations including those of the Abidjan coastal region recognized as major producers and consumers. Attiéké is the main source of income generating activity [6]. Thus, the annual production of fresh attiéké is estimated between 18,965 tons and 40,000 tons. Annual consumption varies between 28 kg and 30 kg per inhabitant [7,8]. Despite its great socio-economic importance, this product has a low nutritional value. To overcome this nutritional problem, strategies for improving local products are put in place. Fortification of complementary foods with other foods rich in micronutrients and macronutrients such as oilseeds and legumes (cashew kernels and moringa leaves) to form a balanced diet is one of the important the fight against malnutrition in children under 5 years old.

In addition, the exceptional nutritional value of oilseeds and legumes (cashew kernel and moringa) is a factor that makes it a potential asset in the fight against malnutrition in Ivory Coast. In fact, oilseeds and legumes contribute to satisfying the need for protein, fiber, lipids, minerals and vitamins, as well as the contribution of functional constituents for the well-being of children. In addition, the exceptional nutritional value of oilseeds and legumes (cashew kernel and moringa) is a factor that makes it a potential asset in the fight against malnutrition in Ivory Coast. In fact, oilseeds and legumes contribute to satisfying the need for protein, fiber, fat, minerals and vitamins, as well as the contribution of functional constituents for the well-being of

children under 5 years old. Thus, they play an essential role in the strategy of children's food security and contribute to nutritional balance. Among oilseeds and legumes, cashew kernels and moringa, which are known to be very nutritious [9], are increasingly used to improve the nutritional content of cereal-based energy foods (corn, rice, millet), tubers (yam, potato) and roots (cassava) low in protein, fat and micronutrients.

In view of all the above, this study aims to produce an infant flour based on attieke enriched with cashew kernel and Moringa powders by determining the physicochemical characteristics and the antioxidant activities in order to contribute to the fight against malnutrition in children under 5 years old.

2. MATERIALS AND METHODS

2.1 Vegetable Material

The Vegetable material used in this study consists of cassava semolina (attiéké) bought in Djahakro, a village located on the outskirts of Yamoussoukro town in three producers, the cashew kernel flour obtained after various treatments of cashew nuts and the powder of the leaves of *Moringa Oleifera*.

2.2 Production of Attiéké Flour

The attieke collected from the producers is dried in an oven at 60°C for 24 hours and crushed using a grinder. The flour obtained was stored in polyethylene bags.

2.3 Production of Deoiled and Unfermented Cashew Kernel Flour

The cashew kernel flour are obtained after dehulling, drying and pruning nuts. The cashew kernel flour is produced according to the method described by [10] modified. The dried cashew kernel are crushed using a semi-artisanal grinder and placed in a stainless steel tank. Hexane is added 1: 1 (w / v) to the flakes for oil extraction. The mixture is macerated for 30 minutes before

being heated at 130°C for 50 minutes and allowed to stand for 24 hours at room temperature. Then the pellet is separated from the supernatant (oil and hexane). The operation is performed twice. The cakes are pressed for 24 hours to extract the rest of the oil. The deoiled cake is oven dried at 70°C for 12 hours. They are milled in a mill and the flour obtained is stored in polyethylene bags.

2.4 Production of Fermented Deoiled Cashew Kernel Flour

The cashew kernels are fermented according to the method of [11] modified. The cashew kernel are boiled at 100°C for 1 hour. The boiled almonds are wrapped in the plantain leaf for 72 hours for fermentation. The fermented seeds are oven dried 60°C for 48 hours. The fermented cashew kernel are crushed and the hexane is added (confers the production of unfermented flour). The cakes are milled in a grinder and packaged in polyethylene bags.

2.5 Production of Moringa Powder

The leaflets of *M. oleifera* leaves are detached from their petioles, sorted to remove damaged leaves and sanitized for 5 minutes in chlorinated water. After rinsing with distilled water and then draining for 30 minutes, the leaflets are soaked for 12 hours and dried out of the sun in an airy room for three weeks and ground in a hammer mill.

2.6 Formulation of Infant Flours: Attiéké / Cashew Kernel/Moringa

Attiéké / cashew kernel / moringa composite flours are obtained by incorporating respective proportions of 10, 15 and 20% moringa flours into the two most preferred composite flours in the study by [12], (FAFCF15: 85% Attiéké flour / 15% fermented cashew kernel, FAFCNF10: 90% Attiéké flour / 10% unfermented cashew kernel). Each formulation is thoroughly mixed in a blender and stored for analysis.

The formulations and their abbreviations

- A : 90% FAFCF15 + 10% moringa
- B : 85% FAFCF15 + 15% moringa
- C : 80% FAFCF15 + 20% moringa
- D : 90% FAFCNF10 + 10% moringa
- E : 85% FAFCNF10 + 15% moringa
- F : 80% FAFCNF10 + 20% moringa

2.7 Physicochemical Parameters of Composite Flours (Attiéké- Cashew Kernel) Enriched with Moringa Powder

The moisture content is determined by the method described in [13] based on oven drying dehydration of samples until a constant weight is obtained. The determination of the ash content is done according to the method [13]. It consists in mineralizing a sample of 5 g (m) at 600°C. for 6 h in a muffle furnace (NABERTERM, GmbH Bahnhofstrasse 20,28865 Lilienthal / Bremen, Germany), until destruction of all organic matter contained in the sample. About 0.1 g of attiéké-cashew kernel composite flour is used to determine the crude protein content from the total nitrogen assay using the Kjeldhal method [13]. The protein level is obtained by multiplying the total nitrogen content by a factor of 6.25 convention. The determination of lipid content is by the soxhlet method [13]. The fat is extracted by boiling with pure hexane. The latter is then removed by evaporation and the residue is dried and weighed. The raw fibers include cellulose, some hemicelluloses and lignin. The raw fiber contents of flours are determined by the method of [14] For this, one gram of flour (m) was boiled in 50 ml of sulfuric acid (1.25 N) and then in 50 ml of sodium hydroxide (1.25 N) for one hour (30 minutes × 2). The resulting residue is dried at 105°C for eight hours (m1) and then incinerated at 550°C for three hours (m2). The total crude fiber (Fb) content, expressed as percentage dry matter.

The determination of carbohydrates is made by difference according to the following formula [13]:

$$\% \text{ Carbohydrates} = 100 - (\% \text{ Moisture} + \% \text{ Protein} + \% \text{ Fat} + \% \text{ Ash}).$$

The energy value is calculated using Atwater specific coefficients:

$$(\% \text{ protein} \times 4) + \% \text{ carbohydrate} \times 4 + \% \text{ lipid} \times 9$$

The total phenols are extracted according to Method [13] (Christensen, 1974), using the Folin Ciocalteu reagent. For extraction, 1 g of flour is placed in a beaker into which are added 100 mL of oxalic acid at 0.3% (g / v). The mixture is stirred with a magnetic stirrer for 30 minutes. The whole is centrifuged at 3000 rpm for 15 minutes. Then the extract is filtered on Whatman paper No. 41. For the assay, 1 mL of the extract diluted

in 8 mL of distilled water is added to a tube, plus 0.5 mL of diluted Folin Ciocalteu reagent (1/10), and 1.5 mL of sodium carbonate solution (7.5%). The mixture is left in the dark for 1 hour at room temperature. The reading is made at an absorption of 765 nm at the spectrophotometer. A standard range is made with gallic acid at different concentrations (0 to 1 mg / mL).

The determination of the antioxidant activity is made according to the method described by [15] whose principle is based on the fading of DPPH. This discoloration is proportional to the antioxidant activity of the sample. A volume of 2.5 mL of methanolic extract is introduced into a test tube. To the contents of the tube is added 1 mL of DPPH solution (3 mM in methanol). The tube is placed in the dark for 30 min and the absorbance is read at 415 nm against the blank. A control tube (1 mL of DPPH + 2.5 mL of methanol) is made and the absorbance of the tube is read under the same conditions as the test tube. The mineral content is determined by atomic absorption spectrophotometry [13]. 0.5 g of ground sample is weighed in a porcelain crucible and then baked at 600°C for six hours. After cooling, 5 ml of 1 mol/L nitric acid are added to the ash obtained and then brought to total evaporation on a sand bath. To the residue are added five milliliters of hydrochloric acid (0.1 mol / L) which is returned to the oven at 400°C for 30 min. The final residue is recovered with 10 mL of hydrochloric acid (1 mol / L) and then poured into a 50 ml flask. The crucible is rinsed twice with 10 ml of hydrochloric acid. The flask is supplemented to 50 ml with hydrochloric acid. The content of the minerals was obtained by air-acetylene flame atomic absorption spectrophotometer assay. The values were read in mg /L. The read values were then converted to mg /100 g.

3. RESULTS AND DISCUSSION

3.1 Physicochemical Properties of Composite Flours (Attiéké- Cashew Kernel) Enriched with Moringa Powder

The various physico-chemical analyzes are presented in Table 1. The pH of the various flours varies from 4.80 ± 0.1 for the sample A and 5.23 ± 0.5 for the moringa powder. Titratable acidity amounts to 10 ± 1.73 meq /100 g (*Moringa oleifera*) and 13.33 ± 0.57 meq / 100 g (A). The addition of *Moringa oleifera* powder increased the moisture content of the Attie-

cashew kernel Composite Flours. However, these values remain below 12% ($7.13 \pm 0.11\%$ at $7.8 \pm 0.2\%$) and all these values do not show significant differences ($P < 0.05$). As for the ash content, they vary from $1.66 \pm 0.57\%$ to $4.33 \pm 0.57\%$.

The fortification of Attiéké-cashew kernel with *Moringa oleifera* composite flour resulted in an increase in the protein content of composite flours. Moringa powder at a content of 24.63 ± 0.10 g /100 g of protein. These contents vary significantly ($P < 0.05$) from 11.59 ± 0.10 g /100 g to 16.59 ± 0.26 g / 100 g. The high protein levels of *Moringa oleifera* leaves have been demonstrated in several studies. The contents of composite flours have increased with the incorporation rate of moringa powder and these values are in line with the standards recommended by [16]. Proteins play a role in the defense of the body and cover the nitrogen expenditure caused by the renewal of tissues and the synthesis of certain compounds involved in the proper functioning of the body (enzymes, hormones) [17].

The fiber contents of the various flours enriched with *Moringa oleifera* are low (2 ± 0.50 g / 100 g - 4.00 ± 0.50 g / 100 g), however the Moringa powder has a high value ($10.66 \pm 1, 50$ g / 100 g) in fibers. The dietary fiber content of food supplements should not exceed 5 g / 100 g of product on a dry weight basis according to [18]. The fibers regulate the intestinal transit; capture some of the fat and carbohydrates and can also reduce the caloric density of complementary food preparations.

The energy values of the different composite flours are statistically different. The Moringa powder has a value of 360.04 ± 1.47 kcal / 100 g, while the energy values of the formulations are between 374.41 ± 1.39 kcal / 100 g and 365.36 ± 1.58 kcal / 100 g.

3.2 Mineral Content of Composite Flours (attiéké- cashew kernel) Enriched with Moringa Powder

Table 2 shows the contents of the different minerals observed in Moringa enriched composite flours. The fortification of Moringa composite flours led to an increase in mineral content. Calcium and potassium are the most abundant in the Moringa powder among the macroelements studied, with respective contents of 1166.68 ± 2 mg / 100 g and 1818.22 ± 0.22

mg / 100 g. For the formulations the calcium contents differ significantly ($P < 0.05$). Flour C has the highest content 394.54 ± 0.5 mg / 100 g and flour D has the lowest value 146.69 ± 0.55 mg / 100 g. The increase in the contents of the various composite mineral flours is due to the incorporation of the moringa powder. Moringa leaves are known for their excellent mineral content [19,20]. As regards the formulations, their calcium contents have increased, however the flours C and F, have satisfactory contents according to the standard [21]. Calcium helps fortify bones, especially in the growing season. It plays a major role in muscle contraction, vitamin B12 absorption and blood clotting [22].

From 15% incorporation of moringa flour the potassium contents are suitable. Potassium acts against disturbances of the heart rhythm and intervenes in the regulation of the osmotic pressure of the cell through the Na^+ / K^+ pump. It also contributes to maintaining the acid-base balance of the body [23]. All formulations have satisfactory levels of magnesium and copper. Copper participates in the synthesis and maintenance of andas myelin [24]. Magnesium is important for the proper functioning of the body, it participates in the cohesion of proteins by activating enzymatic functions, prevention of muscle degeneration, growth retardation and congenital malformations [25]. Iron and zinc are important for the functioning of the body as their deficit poses real public health problems. Zinc is an essential component of many enzymes involved in the synthesis and degradation of proteins, lipids and carbohydrates, in the synthesis of prostaglandins and in the metabolism of other micronutrients [26]. In the human body, although present in very small quantities; iron plays vital roles in vital functions. It is also involved in the formation of hemoglobin, in its heme form, myoglobin and enzymes that play a key role in many metabolic reactions, allowing the transport of oxygen from the lungs to the tissues [27]. Moringa leaves, like leafy vegetables, can help fight micronutrient deficiencies to ensure growth for children under five.

3.3 Antioxidant Property of Composite Flours (Attiéké- Cashew Kernel) Enriched with Moringa Powder

Table 3 shows the contents of vitamin C, phenolic compound and antioxidant activity.

Moringa powder is a major source of total polyphenols 869.59 ± 0.3 mgEAG / 100 g, as well as tannins 232.04 ± 1.06 mg / 100 g. The compound flours have contents of between 434.78 ± 0.1 mgEAG / 100 g for flour D and 609.31 ± 1.22 mgEAG / 100 g for flour C total polyphenols. For tannin, flour F was 147.99 ± 1.26 mg / 100 g the highest statistically ($P < 0.05$) of composite flours. Phenolic compounds are important antioxidants that protect biological macromolecules against degradation [28]. Thus, they effectively fight against aging and the occurrence of cancer cells [29,30]. *Moringa oleifera* powder flour is an important source of total polyphenols, which leads to an increase in these contents in moringa-enriched composite flours.

The vitamin C contents of the different formulations show significant differences ($P < 0.05$). Flour F at the highest content of the formulations 13.98 ± 0.74 mg / 100 g. However, these values are lower than that of Moringa powder 19.81 ± 0.14 mg / 100 g. The analysis of the ability to trap the free radical of 2,2-diphenyl-1-picrylhydrazyl (DPPH) of the various flours to show that the Moringa powder has a significant capacity to trap free radicals with a content of 141.47 ± 0.2 g / 100 g. The antioxidant activity levels of the various flours show significant differences. They range from 71.67 ± 0.22 g / 100 g (D) to 86.94 ± 0.2 g / 100 g (C).

Several authors have also reported that *Moringa oleifera* leaves are known to be an excellent source of antioxidants and a significantly higher content compared to other fruits such as strawberries known for their antioxidant content [31,32]. The antioxidant properties of the methanolic extracts, although variable, indicate that the flours studied are inhibitors of free radicals. The antioxidant efficacy of polyphenols is mainly due to the ease with which a hydrogen atom of an aromatic hydroxyl group is ceded to a free radical [33]. Furthermore, the affinity of polyphenols for free radicals makes it possible to inhibit the oxidation of low-density lipoproteins, playing a positive role in the prevention of cardiovascular diseases [34]. This antioxidant property is beneficial and helps prevent carcinogenesis [35]. A diet based on these plants, in particular, *Moringa oleifera* with high polyphenolic levels could strengthen the health of populations.

Table 1. Results of the physicochemical properties of attiéké / cashew kernel composite flours enriched with *Moringa oleifera* powder

Paramètres	A	B	C	D	E	F	M
pH	4.80±0.1 ^e	4.89±0.01 ^{cd}	4.93±0.05 ^b	4.88±0.3 ^d	4.90±0.2 ^c	4.94±0.25 ^b	5.23±0.5 ^a
Ac. Titrable méq/100 g	13.33±0.57 ^a	11.33±0.5 ^{ab}	10.66±0.55 ^{ab}	12±2 ^{ab}	11.66±1.5 ^{ab}	10.66±1.8 ^{ab}	10±1.73 ^b
Moisture %	7,8±0.2 ^a	7.6±0 ^a	7.2±0.2 ^a	7.6±0.11 ^a	7.5±0.11 ^a	7.6±0.2 ^a	7.13±0.11 ^a
Ash %	2.33±0.57 ^{bc}	2.66±0.57 ^{bc}	3.66±0.50 ^b	1.66±0.57 ^{cd}	2.66±0.20 ^{bc}	3.33±0.30 ^b	4.33±0.57 ^a
Protein %	12.87±0.30 ^d	14.20±0.50 ^c	16.59±0.26 ^b	11.59±0.10 ^e	12.86±0.10 ^d	14.96±0.20 ^c	24.63±0.10 ^a
Fiber %	2±0.50 ^{ef}	2.33±0.28 ^{def}	3.16±0.28 ^{bcd}	3.00±0.50 ^{cd}	3.3±0.50 ^{bc}	4.00±0.50 ^b	10.66±1.50 ^a
Fat %	2.35±0.05 ^a	2.02±0.02 ^a	1.86±0.02 ^{ab}	2.05±0.01 ^a	2.01±0.01 ^a	1.76±0.03 ^{ab}	1.20±0 ^{ab}
Carbohydrate %	78.64±0.61 ^a	77.51±0.2 ^b	73.77±0.80 ^d	79.02±0.58 ^a	77.25±0.4 ^b	76.01±0.5 ^c	62.70±0.61 ^e
Energy kcal/100g	374.41±1.39 ^a	373.05±1.42 ^{ab}	365.36±1.58 ^c	368.91±1.57 ^{bc}	366.58±0.4 ^c	367.76±1.17 ^c	360.04±1.47 ^d

Averages with different letters in the same row are significantly different ($P < 0.05$) according to Duncan's test

A: 90% FAFCF15 / 10% Moringa; B: 85% FAFCF15 / 15% Moringa; C: 80% FAFCF15 / 20% Moringa; D: 90% FAFCNF10 / 10% Moringa; E: 85% FAFCNF10 / 15% Moringa;

F: 80% FAFCNF10 / 20% Moringa; M: Poudre de Moringa

Table 2. Mineral content of attiéke-cashew kernel composite flours fortified with moringa powder (mg / 100 g)

Echantillons	Calcium	Potassium	Sodium	Magnésium	Cooper	Iron	Manganèse	Zinc
A	154.54±1.1 ^f	274±0.4 ^f	12.43±0.25 ^f	92.07±1 ^f	4.6±0.15 ^c	3.41±0.2 ^c	1.45±0.35 ^c	2.60±0.4 ^f
B	231.43±0.35 ^d	416.03±1.52 ^d	14.98±0.4 ^{de}	125.17±1.5 ^d	5.65±0.26 ^c	3.84±0.1 ^c	1.55±0.36 ^c	3.63±0.3 ^{de}
C	394.54±0.5 ^b	588.38±0.8 ^b	16.39±1.21 ^b	166.56±0.39 ^b	8.5±1.00 ^b	5.36±0.85 ^b	3.22±0.25 ^b	4.40±0.25 ^{cd}
D	146.69±0.55 ^g	246.01±0.35 ^g	12.84±0.5 ^f	88.08±0.83 ^g	4.6±0.35 ^c	3.93±0.81 ^c	0.90±0.1 ^c	3.00±0.5 ^{ef}
E	223.10±2.1 ^e	399.04±0.8 ^e	14.4±0.2 ^e	123.23±1.52 ^e	5.82±0.75 ^c	4.59±0.57 ^{bc}	2.58±0.2 ^b	3.81±0.41 ^{cd}
F	353.78±0.6 ^c	512.12±1.32 ^c	15.71±0.25 ^{cd}	159.26±0.37 ^c	7.99±0.86 ^b	5.87±1 ^b	2.96±0.72 ^b	5.04±0.71 ^b
M	1166.68±2 ^a	1818.22±0.22 ^a	66.11±0.2 ^a	523.9±1.6 ^a	33.53±1.53 ^a	28.81±1.2 ^a	4.27±0.35 ^a	14.18±0.2 ^a
Standards (mg / 100 g) FAO/OMS (2006)	341.2	408.7	60	48.7	0.1	8.5	-	3.7

Averages with different letters in the same row are significantly different ($P < 0.05$) according to Duncan's test

A: 90% FAFCF15 / 10% Moringa; B: 85% FAFCF15 / 15% Moringa; C: 80% FAFCF15 / 20% Moringa; D: 90% FAFCNF10 / 10% Moringa; E: 85% FAFCNF10 / 15% Moringa;

F: 80% FAFCNF10 / 20% Moringa; M: Poudre de feuille Moringa

Table 3. Antioxidant properties of composite flours fortified with moringa powder

Sample	Vit C mg/100 g	Polyphenols mgEAG/100 g	Flavonoids MgEAG/100 g	Tannins mg/100 g	Ac. antioxidant mg/100 g
A	11.74±0.43 ^a	471.01±1.75 ^{ab}	88.56±1.48 ^{ab}	100.08±1.98 ^a	76.35±0.3 ^b
B	12.65±0.14 ^b	543.48±0.8 ^{bc}	102.65±1.04 ^{cd}	131.05±1.8 ^b	80.41±0.5 ^d
C	13.48±0.42 ^{cd}	609.31±1.22 ^c	121.46±1.89 ^e	143.49±1.33 ^{bc}	86.94±0.2 ^f
D	11.74±0.45 ^a	434.78±0.1 ^a	82.52±1.96 ^a	104.24±2.88 ^a	71.67±0.22 ^a
E	12.73±0.42 ^{bc}	471.01±1.51 ^{ab}	94.59±0.48 ^{bc}	111.44±1.45 ^a	78.88±0.11 ^c
F	13.98±0.74 ^d	537.63±0.25 ^{bc}	110.70±1.97 ^d	147.99±1.26 ^c	81.29±0.34 ^e
M	19.81±0.14 ^f	869.59±0.3 ^d	221.02±0.11 ^f	232.04±1.06 ^d	141.47±0.2 ^g

Averages with different letters in the same row are significantly different ($P < 0.05$) according to Duncan's test.

A : 90% FAFCF15 / 10% Moringa ; B : 85%FAFCF15 / 15% Moringa; C : 80%FAFCF15 / 20% Moringa; D : 90%FAFCNF10 / 10% Moringa ; E : 85%FAFCNF10 / 15% Moringa; F : 80%FAFCNF10 / 20% Moringa; M: Poudre de feuille Moringa

4. CONCLUSION

The fortification of composite flours (attiéké / unfermented cashew kernel and attiéké / fermented cashew kernel) with the different proportions (10%, 15%, 20%) of *Moringa oleifera* allowed an increase in protein contents (11.59% - 16.59%). The contents of minerals, phenolic compounds and oxidative activities have also increased. *Moringa oleifera* powder would be a good source of fortification for food for children.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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